Agilent - Life Sciences/Chemical Analysis Online Store

Exhibit B

Page 1 of 2

Agilent Technologies

| About Agilent | Products & Services | Industries | International | Online Stores |

Home > Products & Services > Life Sciences/Chemical Analysis > Columns & Accessories > Life Sciences/Chemical Analysis Online Store

Hello
If you are not , click here

Life Sciences/Chemical Analysis Online Store

| Library |
| Technical Support |
| Online Store |
| Education |
| Events |
| News Releases |
| Alliances |
| My Profile |

**Stay Current
With e-notes**

What is Gas Chromatography?

Chromatography is the separation of a mixture of compounds (solutes) into separate components. By separating the sample into individual components, it is easier to identify (qualitate) and measure the amount (quantitate) of the various sample components. There are numerous chromatographic techniques and corresponding instruments. Gas chromatography (GC) is one of these techniques. It is estimated that 10-20% of the known compounds can be analyzed by GC. To be suitable for GC analysis, a compound must have sufficient volatility and thermal stability. If all or some of a compound's molecules are in the gas or vapor phase at 400-450°C or below, and they do not decompose at these temperatures, the compound can probably be analyzed by GC.

The main parts of a basic GC system are shown in Figure 1. One or more high purity gases are supplied to the GC. One of the gases (called the carrier gas) flows into the injector, through the column and then into the detector. A sample is introduced into the injector usually with a syringe or an exterior sampling device. The injector is usually heated to 150-250°C which causes the volatile sample solutes to vaporize. The vaporized solutes are transported into the column by the carrier gas. The column is maintained in a temperature controlled oven. The solutes travel through the column at a rate primarily determined by their physical properties, and the temperature and composition of the column. The various solutes travel through the column at different rates. The fastest moving solute exits (elutes) the column first then is followed by the remaining solutes in corresponding order. As each solute elutes from the column, it enters the heated detector. An electronic signal is generated upon interaction of the solute with the detector. The size of the signal is recorded by a data system and is plotted against elapsed time to produce a chromatogram.

Figure 1. The Basic Components of a GC System
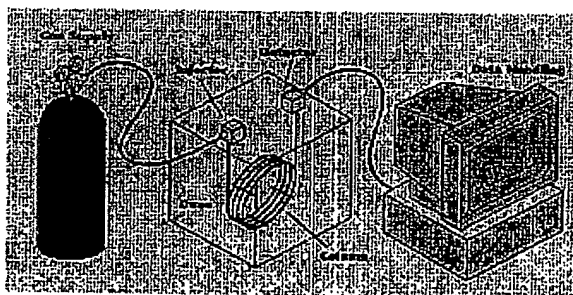
The ideal chromatogram has closely spaced peaks with no overlap of the peaks. Any peaks that overlap are called coeluting. The time and size of a peak are important in that they are used to identify and measure the amount of the compound

in the sample. The size of the resulting peak corresponds to the amount of the compound in the sample. A larger peak is obtained as the concentration of the corresponding compound increases. If the column and all of operating conditions are kept the same, a given compound always travels through the column at the same rate. Thus, a compound can be identified by the time required for it to travel through the column (called the retention time). The identity of a compound cannot be determined solely by its retention time. A known amount of an authentic, pure sample of the compound has to be analyzed and its retention time and peak size determined. This value can be compared to the results from an unknown sample to determine whether the target compound is present (by comparing retention times) and its amount (by comparing peak sizes). If any of the peaks overlap, accurate measurement of these peaks is not possible. If two peaks have the same retention time, accurate identification is not possible. Thus, it is desirable to have no peak overlap or co-elution.

Inside a Capillary GC Column

capillary GC column is comprised of two major parts - tubing and stationary phase. A thin film (0.1-10.0 µm) of a high molecular weight, thermally stable polymer is coated onto the inner wall of small diameter (0.05-0.53 mm I.D.) tubing. This polymer coating is called the stationary phase. Gas flows through the tubing and is called the carrier gas or mobile phase.

Upon introduction into the column, solute molecules distribute between the stationary and mobile phases. The molecules in the mobile phase are carried down the column; the molecules in the stationary phase are temporarily immobile and do not move down the column. As the molecules in the mobile phase move through the column, some of them eventually collide with and re-enter the stationary phase. During the same time span, some of the solute molecules leave the stationary phase and enter the mobile phase. This occurs thousands of times for each solute molecule as it passes through the column. All of the molecules corresponding to a specific compound travel through the column at nearly the same rate and appear as a band of molecules (called the sample band). The goal is to have no overlap between adjacent sample bands as they exit the column. This is accomplished by making each sample band travel at a different rate and by minimizing the width of the sample band. The rate at which each sample band moves through the column depends on the structure of the compound, the chemical structure of the stationary phase and the column temperature. The width of the sample band depends on the operating conditions and the dimensions of the column. The proper column and operating conditions are critical in obtaining no, or the least amount of, peak co-elution.